Keratin Expression during Neoplastic Progression of Bladder Cancer

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Antibodies to keratin proteins can be used to study different cell layers in normal human transitional epithelium, and in human transitional cell carcinomas. Conventional rabbit antisera to human skin keratins stain all layers of the transitional epithelium from bladder, ureter and kidney. A slightly higher staining intensity is found in the basal and superficial layers as compared to the intermediate cell layers. A monoclonal antibody to cytokeratin-18 (RGE 53), however, stains only the superficial cell layer of transitional epithelium, the so-called umbrella cells. Also in well-differentiated (grade-I) transitional cell carcinomas, RGE 53 stains only the superficial cells of the papillary structures. In higher grade papillary tumors, RGE 53 also reacts with cells within the basal and intermediate layers, while in high grade, invasive tumors almost all tumor cells were RGE 53-positive. In a metastatic transitional cell carcinoma all tumor cells are reactive for RGE 53 [Ramaekers et al., 1985].

Therefore it seems that expression of certain cytokeratin-18 epitopes in transitional cell carcinomas changes with an increase in the degree of malignancy of the tumors [Feitz et al., 1985].

For a quantitative analysis of this phenomenon we have analyzed transitional cell carcinomas by flow cytometry (FCM) using propidium iodide for DNA analysis and antibodies to cytokeratin by indirect immunofluorescence. By means of two-dimensional FCM analysis, cytokeratin-positive tumor cells can be analyzed separately from cytokeratin-negative stromal and inflammatory cells. This results in an 18% increase in sensitivity of FCM detection of aneuploidy. In addition, S-phase can be determined in aneuploid samples by means of this two-parameter analysis, where it is not possible using

Fig. 1. a Correlation between the percentage of cytokeratin-positive tumor cells in S-phase and histologic grade of transitional cell carcinoma. Mean GI, 3.4 (s.d. = 1.83); GII, 7.51 (s.d. = 2.98), and GIII, 15.04 (s.d. = 8.72). Wilcoxon test for degree of significance: GI-GII, p = 0.02; GI-GIII, p = 0.014, and GII-GIII, p = 0.014. The horizontal bars indicate the mean values. b Histogram showing the relation between tumor grade, ploidy and percentage of cells positive for RGE 53 (cytokeratin-18) as related to the total number of epithelial tumor cells, estimated by labelling with pKer. Mean GI, 0.02 (s.d. = 0.014); GII, 0.165 (s.d. = 0.113), and GIII, 0.443 (s.d. = 0.335). Wilcoxon test for the degree of significance: GI-GII, p = 0.029; GI-GIII, p = 0.018, and GII-GIII, p = 0.107. x = Diploid sample; o = aneuploid sample. Horizontal bars indicate the mean values.
only DNA content because of the overlap of diploid aneuploid populations [Smeets et al., 1987a, 1987b].

FCM analysis allows quantification of the percentage of tumor cells expressing cytokeratin-18 which can be shown to correlate quantitatively with higher grade, higher stage transitional cell carcinomas (fig. 1).

The quantitative measurement of tumor cell expression of cytokeratin-18 by FCM analysis appears to provide additional information of potential prognostic value independent of tumor cell ploidy and proliferative fractions.

References


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